Euglena Viridis (Ehrenberg.)

By Sister M. Ellen.

This flagellate was found in unusual abundance early in January, 1921, in a little spring-fed creek which remained open all winter. This creek is the watering trough for cattle and other farm animals and the water is more or less polluted as seems to be the characteristic condition under which *Euglena viridis* grows in abundance. About the middle of January, it was observed that the bottom of this creek was completely lined with the rather gelatinous, green masses of Euglena cells. This condition continued all winter and, at the suggestion of Dr. Bert Cunningham, the following investigations were made:

Test of Evolving Gas

On April 12, much inflated masses of the Euglena cells were floating on the surface of the water. A quantity of the material was collected and a test of the evolving gas proved it to be oxygen. Masses of the material, especially after they had stood in the laboratory for a few hours, invariably emitted a decidedly 'fishy' odor. Such an odor is mentioned by Butschli in describing *E. sanguinea.*

Tests for Chlorophyll

After a quantity of the material had stood for several hours in a large stender dish exposed to the light, the thick, green, oily layer of Euglena cells was poured off and allowed to filter, the cells remaining on the filter paper. The filter paper was then transferred to a beaker of methyl alcohol which was heated to the boiling point over a water bath.

In order to check the results of the test another alcoholic extraction was prepared in the same way from fresh parsley leaves. After the extraction from the Euglena cells was complete, the green liquid was put through a filter, leaving the cells on the filter paper. These, when examined under the microscope, showed practically none of

their original color, but were intact and but little shrunken by this treatment. The separation of the pigments of the chlorophyll extractions, both of the Euglena cells and of the parsley leaves was done by adding to about 20 c. c. of each of the extracts the same amount of benzine. The vials were then shaken vigorously and their contents allowed to stand until the pigments separated. The yellow alcoholic layer and the blue-green benzine layer were identical in the two extracts. The flourescence, too, both of the chlorophyll solutions and of the cyanophyll-benzine layers were the same in the Euglena and the parsley extractions. Tests for the effect of sunlight and of darkness on both extracts gave corresponding results.

Some of the Euglena cells were boiled in water for a minute or so. No pigment of any kind discolored the water and the cells themselves, when examined under the microscope, showed no color change and but little distortion or shrinkage. It was observed, however, in the cells subjected to this treatment, that the flagella were made quite prominent.

Cultures

Such media as tap water,* creek water, (filtered and unfiltered) Marschal's solution, Byrenck's solution, very dilute solutions of cane sugar, prune juice, egg white, egg yolk, whole egg, and milk, all tried in darkness and in light under the same conditions of temperature, gave results, that is, motility, growth, and reproduction, incomparably better in the light than in the darkness. The best results obtained from the inorganic nutrients, creek water, and tap water, in moderate light. These results were all about equally good. The second best results were in the organic nutrients in the light. The cultures which ranked as a distant third were those in the organic nutrients in the dark. In these, a comparatively few cells divided longitudinally after a few days, but there was little motility. The chloroplasts in these cells, in general, did not disintegrate any more rapidly than did the cells themselves. A very few motile cells showed a number of non-green plastids.

Among the hindrances in the cultural work, are the numerous protozoan forms which thrive in the cultures about equally well in light and darkness such as species of Paramoecium, Vorticella, Stylonychia,

* This is deep well water which, on chemical analysis, yields calcium, iron, magnesium, the hydroxyl, sulphate, and carbonate radicals, and a trace of chlorine.

Dileptus, Amoeba, etc. Numerous Rotifera, too, together with the protozoa, decrease the cultures by feeding on the Euglena cells. Protozoa as well as bacteria were rife in the organic nutrients but not in the inorganic nutrients nor in the creek water and tap water.

While the encysted cells are still alive, the water mould, *Polyphagus Euglenae* grows rapidly in the cultures, especially so in the unfiltered creek water. The growth and decline of this mould are an index on the condition of the encysted Euglena cells. Thus, it declines in the cultures in the darkness sooner than in the light. It was observed that, like some of the other water moulds such as Saprolegnia, the zoospores of *Polyphagus Euglenae* swarm only in the morning.

Reproduction

Reproduction in the successful cultures begins about the second or the third day and, at first, there is only longitudinal division, the parent cells coming to rest at night, and the daughter cells separating in the morning. As the cultures (in light) advance, many of the cells encyst and there arises from each encystment, four, eight, and even sixteen cells; four being the most common number. There are many eight-celled encystments but relatively few sixteen-celled groups.

Summary

1. The discharge of oxygen gas from masses of Euglena cells in the sunlight indicates the process of photosynthesis.

2. The green plastids of *Euglena viridis* are chlorophyll-bearing.

3. *Euglena viridis* does not thrive in the dark and it, therefore, must depend, at least very largely, upon photosynthesis for its nutrition. There is an indication, however, that *Euglena viridis* does make use of organic nutrients to a very slight degree, in the fact that it seems to be a little more successful in such media when in the dark than in inorganic nutrients. This may be accounted for in part, however, by the fact that the protozoa and other enemies of Euglena are less dependent upon it in organic media than they are in the inorganic media.

4. Reproduction in *Euglena viridis,* as observed in this study, is by longitudinal fission of resting cells, and by the division of encysted cells into four, eight, or even sixteen cells.

Saint Clara College,
Sinsinawa, Wisconsin.